AMENDMENTS TO THE SPECIFICATION

Please replace paragraph [0007] on page 2 with the following rewritten paragraph:

-- A device and method of the present invention allows evaluation of the contents of a sealed primary container by means of an integral sensor which is separated from the contents of the sealed primary container yet provides information on quality of the contents of the primary container without breaking the sealed system. The integral sensor device can be sealed into the seam of a blood bag or seam of another type of primary container. The integral sensor device includes a biosensor retained within a plastic construct by a gated-pore membrane. Pores in the membrane open in response to an environmental change in the primary container allowing the contents of the primary container to contact the biosensor. Status of the contents of the primary container can be determined by inspection of the biosensor, visually or via a fiber-optic probe, through the optical window of the plastic construct. --

Please add the following new paragraph after paragraph [0011] on page 3:

-- Fig. 4a is a schematic representation of a device according to the present invention, which has been sealed in the seam of a blood bag, shown as a cut-away from the blood bag as a whole. --

Please add the following new paragraph after paragraph [0012] on page 3:

-- Fig. 5a is a schematic representation of a device according to the present invention, which has been sealed in the seam of a blood bag, shown as a cut-away from the blood bag as a whole. --

Please replace paragraph [0020] on page 3 with the following rewritten paragraph:

-- Integral sensor – In this disclosure, we use the term "integral sensor" to designate one of many devices as disclosed herein. An "integral sensor" typically is incorporated into a primary container, prior to sterilization if appropriate, and separates a biosensor (see below) from the contents of the sealed primary container yet provides information on quality of the contents of the container without breaking the sealed system. In a preferred embodiment, the "integral sensor" or "sensor device" is incorporated into the seam of a blood bag such that the gated-pore membrane of the integral sensor is exposed to the interior portion of the blood bag. --

Please replace paragraph [0027] on page 5 with the following replacement paragraph:

-- The present invention consists of devices and methods which allow evaluation of the contents of a sealed primary container by means of an integral sensor which is separated from the contents of the sealed primary container yet provides information on quality of the contents of the primary container without breaking the sealed system. The integral sensor device might be a hollow cylinder or a shallow construct, which can be sealed into the seam of a primary container, i.e., blood bag. One end or face of the device is a gated-pore membrane whose pores normally are occluded, by one of many approaches, forming one end of a sensor compartment, containing a biosensor appropriate for the task, with the other end of the sensor compartment formed by an optical window recessed in from the end opposite to the gated-pore membrane or formed by a wall of the primary container. Typically, the integral sensor device is fabricated separately from the primary container and incorporated into the primary container during final fabrication, before sterilization. ~~Certain embodiments of the device are therefore capable of aseptic operation. The status of the~~

~~contents of a primary container for blood cells, other cells, foods, or industrial products can be determined by inspection, visually or via a fiber optic probe through the optical window of a plastic construct incorporated into said primary container at fabrication, of the biosensor retained within said plastic construct by a gated pore membrane, the pores in which opened in response to an environmental change in said primary container allowing the contents of said primary container to contact and cause a change in said biosensor. In one embodiment, after the cell suspension or product is placed into the container, the gated pore membrane opens and allows continuous exposure of the biosensor, within the sensor compartment, to the contents of the primary container. Changes within the primary container affect the signal from the biosensor, which can be quantified at any instant in time by viewing the optical window of the device with an appropriate sensor, as above, providing a measure of quality without opening the primary container. In another embodiment, after the cell suspension or product is placed into the container, the gated pore membrane remains closed until a predetermined change occurs within the primary container which causes the gated pore membrane to open allowing fluid to enter the sensor compartment and contact the biosensor, which then responds with an appropriate signal. Changes in the primary container that can be detected include, but are not limited to, a decrease or increase in pH away from a threshold value or accumulation of one or more members of a preselected class of molecules, including toxins produced by bacteria, above a threshold value. A great range in utility is possible because, depending on the device and method, both the material(s) occluding the gated pore membrane and material(s) forming the biosensor can be varied independently or in combination. Hence, a predetermined change in contents of the primary container can be evidenced by opening of the gated pore membrane and/or a change in the signal from the biosensor. A number of other embodiments or uses would be obvious to one skilled in the art, and the examples herein illustrate but do not set the limits of this invention.~~ --

Please add the following new paragraphs after paragraph [0027] on page 5:

-- In a preferred embodiment, the integral sensor is incorporated in the seam of a blood bag. The integral sensor is sealed with a sealant known in the art to create an impervious seal between the integral sensor and the seam of the blood bag, which is created by sealing two pieces of plastic or other composite known in the art together. Once incorporated into the seam of the blood bag, the face of the integral sensor having the gated-pore membrane is exposed to the interior of the blood bag for exposure to the contents of the bag and the optical window of the integral sensor is exposed to the environment exterior to the blood bag. --

-- Certain embodiments of the device are therefore capable of aseptic operation. The status of the contents of a primary container for blood cells, other cells, foods, or industrial products can be determined by inspection, visually or via a fiber-optic probe through the optical window of a plastic construct incorporated into said primary container at fabrication, of the biosensor retained within said plastic construct by a gated-pore membrane, the pores in which opened in response to an environmental change in said primary container allowing the contents of said primary container to contact and cause a change in said biosensor. In one embodiment, after the cell suspension or product is placed into the container, the gated-pore membrane opens and allows continuous exposure of the biosensor, within the sensor compartment, to the contents of the primary container. Changes within the primary container affect the signal from the biosensor, which can be quantified at any instant in time by viewing the optical window of the device with an appropriate sensor, as above, providing a measure of quality without opening the primary container. In another embodiment, after the cell suspension or product is placed into the container, the gated-pore membrane remains closed until a predetermined change occurs within the primary container which causes the gated-pore membrane to open allowing fluid to enter the sensor

compartment and contact the biosensor, which then responds with an appropriate signal. Changes in the primary container that can be detected include, but are not limited to, a decrease or increase in pH away from a threshold value or accumulation of one or more members of a preselected class of molecules, including toxins produced by bacteria, above a threshold value. A great range in utility is possible because, depending on the device and method, both the material(s) occluding the gated-pore membrane and material(s) forming the biosensor can be varied independently or in combination. Hence, a predetermined change in contents of the primary container can be evidenced by opening of the gated-pore membrane and/or a change in the signal from the biosensor. A number of other embodiments or uses would be obvious to one skilled in the art, and the examples herein illustrate but do not set the limits of this invention. --

Please replace paragraph [0050] on page 16 with the following rewritten paragraph:

The Examples illustrate how biosensors, as above, can be positioned in front of an optical window, accessible to a removable fiber-optic detector, while protected in a separate closed sub-container <u>incorporated</u> within the primary container. They are illustrative of the many uses of the invention, although the invention encompasses other biosensors, other membranes with open pores or gated-pores, other configurations, and other combinations of functional elements in the device. Examples of devices include reference to welding of a gated-pore membrane to a plastic construct. The invention is not limited by the plastic(s) used to form the device. The preferred welding process will depend on the plastic used to fabricate the construct, and the invention includes any method to assemble the device or incorporate it into the primary container, such as ultrasonic and radio-frequency welding <u>sealing the device into a seam of a primary container,</u> or other methods known to those skilled in the art. Examples illustrate the variety of materials that can be used to form

gated-pore membranes or serve as biosensors, in one or more types of devices, and the invention is not limited in respect to specific compositions or analytes.

Please replace paragraph [0052] on pages 17-19 with the following rewritten paragraph:

The following list describes the Figs. in greater detail:

Fig. 1 Schematic representation of a gated-pore membrane prepared using a microporous or capillary-track membrane and unique material to occlude the pores (e.g., as in U.S. Patent No. 5,026,342).

Fig. 2 Schematic representation of changes in a receptor-fluorescent reporter group upon change from unoccupied receptor to occupied receptor-ligand complex, listed by biosensor class as detailed in the specification. A reporter group that is non-fluorescent is designated with a lower-case letter and a reporter group that can fluoresce when excited with light of appropriate wavelength is designated with an upper-case letter.

Fig. 3 Cross-section through a simple boat-shaped device as described in Example 1. The capillary-pore membrane (302) was processed with material (303) to plug the pores and make a gated-pore membrane (304). This membrane is bonded to a plastic construct (301) to form a cavity which then was filled with a biosensor (305) before the device was bonded via a flange (301a) to the primary container (306).

Fig. 4 <u>and 4a</u> Longitudinal section through a device, and also a top view, as described in Example 2. The biosensor-compartment in a cylindrical construct (411) was filled with a biosensor (405). A gated-pore membrane (404) was bonded to a recessed surface (411a) to seal the biosensor in the device. The device then can be incorporated into the seam <u>(420)</u> of a primary container ~~(not shown)~~

(421) as shown in Fig. 4a so that the optical window (412) can be interrogated with a fiber-optic probe from outside the primary container (421), without interrupting the airtight seal, and so that the face of the integral device is exposed to the interior storage compartment of the primary container (421).

Fig. 5 and 5a Longitudinal section through a device, and also a top view, as described in Example 3. An optically clear cylindrical construct (511) was surrounded by an opaque outer concentric construct (515). The biosensor compartment was filled with a biosensor (505). A gated-pore membrane (504) was bonded to a recessed surface (515a) of the opaque outer concentric construct (515) to seal the biosensor in the device. The device then can be incorporated into the seam (520) of a primary container ~~(not shown)~~ (521) as shown in Fig. 5a so that the optical window (512) can be interrogated with a fiber-optic probe from outside the primary container (521), without interrupting the airtight seal, and so that the face of the integral device is exposed to the interior storage compartment of the primary container (521).

Fig. 6 Intensity of fluorescence from a solution of fluorescein isothiocyanate (FITC) in a device as a function of concentration of FITC. Device as in Fig. 5; measurements with fiber-optic detector using excitation at 480 nm and detection at 535 nm (thousands of fluorescent units).

Fig. 7 Intensity of fluorescence from beads in a device as a function of the ratio of beads labeled with fluorescein isothiocyanate (FITC) to plain beads. Device as in Fig. 5; measurements with fiber-optic detector using excitation at 480 nm and detection at 535 nm (thousands of fluorescent units).

Fig.8 Intensity of fluorescence from beads in a device as a function of their known content of fluorescein isothiocyanate (FITC). Known contents of FITC were

equivalent to 0.00, 0.22 and 1.20 million molecules of FITC in solution. Device as in Fig. 5; measurements with fiber-optic detector using excitation at 480 nm and detection at 535 nm (millions of fluorescent units).

Fig. 9 Intensity of fluorescence from beads, previously labeled with fluorescein isothiocyanate (FITC), in a device as a function of pH of the salts solution around the device and around the beads therein. Device as in Fig. 5; measurements with fiber-optic detector using excitation at 480 nm and detection at 535 nm (thousands of fluorescent units).

Fig. 10 Minimal effect of pH of a salts solution on fluorescence from phycoerythrin-cyanin-5 strepavidin attached to beads. Device as in Fig. 5; measurements with fiber-optic detector using excitation at 480 nm and detection at ≥640 nm (thousands of fluorescent units).

Fig. 11 Ratio of emissions from beads responsive to pH and non-responsive to pH can be used to normalize data for irregularities of response or color of the medium. The device contained a mixture of beads labeled with fluorescein isothiocyanate (pH responsive; Fig. 9) and phycoerythrin-cyanin-5 strepavidin (pH non-responsive; Fig. 10). The ratio of emission at 535 nm and ≥640 nm was calculated as a function of pH of the salts solution around the device and around the beads therein. Device as in Fig. 5; measurements with fiber-optic detector using excitation at 480 nm and detection at 535 nm and ≥640 nm (raw values ranged between 75,000 and 135,000 relative fluorescent units). –

Please replace paragraph [0054] on pages 19-20 with the following rewritten paragraph:

-- Another device (Fig. 4) can be used to monitor one of a variety of changes within a closed primary container, when visual detection of changes in the biosensor or the configuration of the device in Example 1 is inappropriate. This device can be of any convenient shape and formed from any plastic, but a cylindrical plastic construct formed from clear acrylic plastic is favored for some applications, and a clear optical window transmitting the portion of the spectrum of interest is essential. The device of Fig. 4 is a cylindrical acrylic construct (411) with a biosensor compartment formed by an optical window (412), a gated-pore membrane (404) responding to any aqueous environment, and the walls of the construct. After the biosensor compartment is filled with a biosensor (405) capable of detecting the desired change within the primary container, the gated-pore membrane is welded to the surface (411a) of a recess in the end of the construct. After the device is incorporated into a primary container, ~~(not shown)~~ shown in Fig. 4a as a cutout section of a blood storage bag with an integral sensor sealed within a seam of the blood storage bag, and the container is filled with a suspension of cells of interest, the biosensor can be interrogated without opening the primary container, when desired, by positioning the tip of a separate fiber-optic probe (not shown), as known to those skilled in the art, against the optical window (412). This device uses the gated-pore membrane simply to protect and isolate the biosensor until the primary container is filled, for example with a cell suspension, after which the gates open in <30 minutes and selectivity in response to a predetermined analyte and intensity of signal are properties of the biosensor and amount of material impinging thereon. --

Please replace paragraph [0055] on page 20 with the following rewritten paragraph:

-- A third device (Fig. 5) incorporates an opaque outer plastic element (515), with accommodation (515a) to weld a gated-pore membrane (504) to this part of the device, positioned around and bonded to an inner element (511) which is a clear plastic construct

similar to the device of Example 2 and contains a biosensor (505) capable of detecting the desired change within the primary container. The device is incorporated into a primary container, shown in Fig. 5a as a cutout section of a blood storage bag having the device sealed within a seam of the blood storage bag. This device can be of any convenient shape and formed from any plastic, but a cylindrical concentric construct is favored for some applications. The outer opaque element (515) might be of colored polyvinyl chloride to shield the biosensor from stray light and facilitate welding the device into a polyvinyl chloride blood bag, and the inner element of clear plastic so that the optical window (512) will transmit the portion of the spectrum of interest. This device can be used to monitor one of a variety of changes within a closed primary container, when stray light might affect detection of the signal from the biosensor, by positioning the tip of a separate fiber-optic probe (not shown), as known to those skilled in the art, against the optical window (512). This device uses a gated-pore membrane to protect and isolate the biosensor until the primary container is filled and for a predetermined interval (e.g., 0.5, 2 or 5 days) thereafter or until a predetermined threshold of a specific change in contents of the primary container is reached (e.g., concentration of first analyte), after which the gates open and the biosensor begins to monitor changes in concentration of the same molecule or ion triggering the gated-pore membrane. Alternatively, the biosensor can be used to monitor concentration of a second analyte with intensity of the signal from the biosensor dependent on properties of the biosensor and amount of second analyte impinging thereon.